Remarks

Reconsideration of this Application is respectfully requested.

Claims 1-30 are pending in the application, with claims 1 and 2 being the independent claims. Based on the following remarks, Applicants respectfully request that the Examiner reconsider the rejections under 35 U.S.C. § 112, first paragraph, and that they be withdrawn.

I. Interview With the Examiner

Applicants thank the Examiner for the courtesy of an interview with the undersigned on May 20, 2003, during which the enablement rejection of claims 5-16 was discussed.

II. Drawings

The drawings were objected to by the draftsperson under 37 C.F.R. § 1.84 or 1.152. (*See* Paper No. 20, page 2.) Applicants thank the Examiner for noting that correction of the defects can be deferred until the application is allowed.

III. Double Patenting

Claims 1-30 were rejected under the judicially-created doctrine of obviousness-type double patenting as allegedly being unpatentable over claims 1-16 of U.S. Patent No. 5,972,634. (*See* Paper No. 20, page 3.) Applicants respectfully request that this rejection be held in abeyance until the remaining issues in the application have been resolved.

IV. Claim Rejections Under 35 U.S.C. § 112, First Paragraph

A. Enablement

Claims 5-16 were rejected under 35 U.S.C. § 112, first paragraph, as allegedly containing subject matter that was not described in the specification in such a way as to enable one skilled in the art to which it pertains, or with which it is most nearly connected, to make and/or use the invention. (*See* Paper No. 20, page 3.) Applicants respectfully traverse this rejection.

Claims 5-16 are directed to diagnostic assays for detecting or quantifying Aβ peptide which may be present in a candidate solution. The assays of claims 5-16 comprise the use of a polyclonal antibody that is specific to $A\beta_{1-42}$ and does not cross-react with $A\beta_{1-40}$ (claims 5, 7, 9, 11, 13 and 15), and a polyclonal antibody that is specific to $A\beta_{1-40}$ and does not cross-react with $A\beta_{1-42}$ (claims 6, 8, 10, 12, 14 and 16) .

According to the Examiner, "[the] specification, as originally filed, fails to teach how to make the claimed antibodies [sic] because [it] lacks an adequate written description of the immunogen to produce such." (Paper No. 20, page 4.) Thus, the rejection is apparently based on the Examiner's position that practicing the claimed assays would have required undue experimentation because the specification does not set forth a particular immunogen that allegedly is required for producing the polyclonal antibodies that are used in the assays of claims 5-16.

The Examiner's position assumes that producing polyclonal antibodies that are specific to $A\beta_{1-42}$ and that do not cross-react with $A\beta_{1-40}$, and vice versa, could only have been accomplished by identifying particular immunogens that are capable of eliciting the

production of such polyclonal antibodies. Although Applicants maintain their position that identifying specific immunogens would have been a matter of routine experimentation in the art, (*see* Applicants' remarks set forth in the Supplemental Amendment and Reply, filed on January 10, 2003, pages 4-7), Applicants further note that other methods -- *not involving the identification of any particular immunogen* -- would have also been available to persons of ordinary skill in the art.

An alternative and well known method for producing specific polyclonal antibodies that does not require the identification of specific immunogens is immunoaffinity purification. There are many variations on immunoaffinity techniques that could have been used to generate polyclonal antibodies that are specific to $A\beta_{1-42}$ and that do not cross-react with $A\beta_{1-40}$, and vice versa. Such techniques had been commonly used in the art for several years prior to the effective filing date of the present application and therefore would not have involved undue experimentation. An outline of one exemplary method is as follows:

- An animal is immunized with the $A\beta_{1-40}$ peptide or fragment thereof, thereby generating a polyclonal response within the animal. The polyclonal antiserum obtained from the animal contains a mixture of antibodies, including antibodies that recognize *only* $A\beta_{1-40}$[1] and antibodies that recognize *both* $A\beta_{1-40}$ and $A\beta_{1-42}$, as well as other non-specific antibodies naturally present in the polyclonal antiserum.
- The polyclonal antiserum is applied to a column containing $A\beta_{1-40}$ peptide immobilized on beads. Two classes of antibodies are retained on the

[1]The polyclonal antiserum will necessarily contain antibodies that recognize *only* $A\beta_{1-40}$ and do not cross-react with $A\beta_{1-40}$ as evidenced by the existence of $A\beta_{1-40}$-specific monoclonal antibodies. (*See, e.g.*, Iwatsubo *et al.*, *Neuron 13*:45-53 (1994).)

column: (1) antibodies that recognize *only* $A\beta_{1-40}$ ("$A\beta_{1-40}$-specific antibodies"); and (2) antibodies that recognize *both* $A\beta_{1-40}$ and $A\beta_{1-42}$ ("cross-reactive antibodies"). The non-specific antibodies are removed in the flow-through.

- The $A\beta_{1-40}$-specific antibodies and the cross-reactive antibodies are eluted from the column and are then applied to a second column containing $A\beta_{1-42}$ peptide immobilized on beads.
- The cross-reactive antibodies will be retained on the column. The flow-through is kept and will necessarily contain only the $A\beta_{1-40}$-specific antibodies.

The general method outlined above, and variations thereof, were well known and commonly practiced by persons of ordinary skill in the art prior to the effective filing date of the application. (*See* discussion immediately below.) In addition, the materials needed to carry out the method (*e.g.*, $A\beta_{1-40}$ and $A\beta_{1-42}$ peptides) were readily available. (*See, e.g.*, specification at page 23, lines 7-17.) Furthermore, specific amino acid sequences which could be used as immunogens are disclosed in the specification. (*See, e.g.*, specification at page 23, lines 16-17, and Fig. 10.)

Selecting antibodies with desired specificities using immunoaffinity techniques is described in the following references: *Practical Immunology*, pp. 192-198, Hudson and Hay, eds., Blackwell Scientific Publications (1976) (copy submitted herewith as Exhibit 1); *Laboratory Techniques in Biochemistry and Molecular Biology*, pp. 466-471, Work and Work, eds., North-Holland Publishing Co. (1979) (copy submitted herewith as Exhibit 2); *Biochemistry*, pp. 789-791, Stryer, ed., W.H. Freeman and Co. (1981) (copy submitted

herewith as Exhibit 3); and *Essential Immunology*, pp. 76-77, Roitt, ed., Blackwell Scientific Publications (1988) (copy submitted herewith as Exhibit 4). These references demonstrate that immunoaffinity purification of antibodies with desired specificities was well established in the art many years prior to the effective filing date of the present application. For example, as stated by Hudson and Hay:

> Besides their use for isolation of pure antibodies, immunoadsorbents are widely used to render antisera specific by *depletion of cross-reacting antibodies* (Section 1.6.6), and for quantitative adsorption (Section 9.1).

(Hudson and Hay, page 198, Section 8.1.5, emphasis added.)

In addition to the above-cited references, there are many examples from the scientific literature describing the use of immunoaffinity techniques to produce polyclonal antibodies that recognize a specific peptide but do not recognize other closely related peptides. In fact, the production of polyclonal antibodies that are specific to $A\beta_{1-42}$ and that do not cross-react with $A\beta_{1-40}$, and vice versa, using immunoaffinity techniques, has been reported. (*See, e.g.*, Mak *et al.*, *Brain Res. 667*:138-142 (1994) ("Mak")) (copy submitted herewith as Exhibit 5). According to the Mak authors:

> Rabbit antisera were raised as previously described, affinity purified on peptide agarose bead columns, and tested against synthetic β40 and β42. Affinity purified antibodies to β37-42 were absorbed on a β40 column and 20 μg/ml of aggregated β40 to remove β-conformation dependent antibodies. The β40 absorbed 37-42 antibody was used in subsequent studies ('anti-42'). The β34-40 antisera from one rabbit ('anti-40') was predominantly reactive against β40 *and was specific for β40 after absorption on β42.*

(Mak at page 138, right column, emphasis added, internal citations omitted.)

Polyclonal antibodies that are specific to $A\beta_{1-42}$ and that do not cross-react with

$A\beta_{1-40}$, and vice versa, are commercially available from Oncogene Research Products, Boston, MA. (*See* Oncogene Research Products catalog, Cat. Nos. PC149 and PC150 (1996)) (copy submitted herewith as Exhibit 6). According to the catalog, the $A\beta_{1-40}$- and $A\beta_{1-42}$-specific polyclonal antibodies were produced using an immunoaffinity procedure similar to that which is outlined above:

> β-Amyloid [1-40] (Ab-1) is a rabbit polyclonal IgG against a C-terminal fragment of β-amyloid peptide [1-40] which has been preabsorbed against both the full length [1-42] and [1-43] followed by affinity purification using a C-terminal fragment of Aβ [1-40].
>
> * * *
>
> β-Amyloid [1-42] (Ab-1) is a rabbit polyclonal IgG against a C-terminal fragment of β-amyloid peptide [1-42] which has been preabsorbed against both the full length [1-40] and [1-43] followed by affinity purification using a C-terminal fragment of Aβ [1-42].

(Oncogene Research Products catalog, page 3.)

The materials and methods used to produce the $A\beta_{1-40}$- and $A\beta_{1-42}$-specific polyclonal antibodies described in Mak and the Oncogene Research Products catalog were well known and available to persons of ordinary skill in the art before the effective filing date of the present application. Thus, the polyclonal antibodies included in the practice of the methods of claims 5-16 could have been produced with only routine experimentation.

Aside from $A\beta_{1-40}$- and $A\beta_{1-42}$-specific polyclonal antibodies, there are other examples in the scientific literature of polyclonal antibodies that recognize a particular peptide but do not cross-react with other closely related peptides. Such polyclonal antibodies were produced using immunoaffinity methods similar to that which is outlined above.

Clément *et al.*, *Circulation Res. 85*:e51-e58 ("Clément") (copy submitted herewith as Exhibit 7), for example, describes the production of polyclonal anti-α-skeletal actin

antibodies that do not cross-react with other actin isoforms, the amino acid sequences of which are closely related to that of the α isoform. (*See* Clément, page e54, Fig. 2.B.) In fact, the polyclonal antibodies produced by the method of Clément were able to distinguish actin peptides that differed from one another by as little as two amino acids. (*See id*, comparing the blocking effects of α-skeletal and α-cardiac actin NH_2-terminal decapeptides.) The method used to produce the specific polyclonal antibodies is set forth on page e52, left column and e53, left column.

The method of Clément involved first producing an antiserum against the NH_2-terminal α-skeletal decapeptide. Next, the antiserum was applied to a column containing Sulfolink beads coupled with the NH_2-terminal α-skeletal decapeptide. Bound antibodies were eluted and then applied sequentially to: (1) a Sulfolink column coupled with the NH_2-terminal decapeptide of β-cytoplasmic actin; and (2) a Sulfolink column coupled with the NH_2-terminal decapeptide of γ-smooth muscle actin, to remove the population of antibodies cross-reacting with these two isoforms. (*See* Clément, page e52, left column.) According to the authors, "[t]he experimental strategy we used here shows that it is possible to produce in the rabbit and immunopurify an antibody that reacts specifically with α-skeletal actin." (*See* Clément, page e55, right column.)

Immunoaffinity methods have even been used to produce polyclonal antibodies that recognize the phosphorylated version of a protein but not the un-phosphorylated version. (*See, e.g.*, Mothe-Satney *et al.*, *J. Biol. Chem. 275*:33836-33843 (2000) ("Mothe-Satney")) (copy submitted herewith as Exhibit 8). In Mothe-Satney, rabbits were first immunized with phosphopeptides corresponding to portions of the translation initiation factor PHAS-I. In order to select for phospho-specific antibodies, the antiserum was incubated with non-

phosphorylated peptides that had been coupled to Sulfolink resin. The fractions that did not bind to the non-phosphorylated peptide resin were incubated with the respective phosphopeptides coupled to Sulfolink. The phospho-specific antibodies were then eluted from the columns following a washing step. (*See* Mothe-Satney, page 33837, left column.) The resulting antibodies were found to recognize the phosphorylated form of PHAS-I but not the unphosphorylated form of the protein. (*See* Mothe-Satney, page 33837, right column.)

The methods described in Clément and Mothe-Satney were available as of the effective filing date of the present application. Therefore, the polyclonal antibodies that are used in the methods of claims 5-16 of the present application could have been produced using methods similar to those described in Clément and Mothe-Satney with only routine experimentation.

Finally, with respect to producing polyclonal antibodies that recognize $A\beta_{1-42}$ but not $A\beta_{1-40}$, the Examiner stated that "[u]sing larger peptides, necessarily encompasses residues in the 1-40 regions and as previously set forth, this immunogen used would produce a polyclonal antibody with a wide range of specificities that include both species." (Paper No. 20, page 7.) Applicants respectfully disagree.

As discussed above, in order to produce polyclonal antibodies that recognize $A\beta_{1-42}$ but not $A\beta_{1-40}$, any cross-reactive antibodies can easily be "subtracted out" by passing the polyclonal antiserum over a column containing $A\beta_{1-40}$ peptide immobilized on beads. This is precisely the method that is described in Mak and the Oncogene catalog.

Furthermore, the specificity of antibodies is based on their interaction with the three dimensional structure of proteins which can vary greatly despite small differences at the

amino acid level. Even though the amino acid sequences of $A\beta_{1-40}$ and $A\beta_{1-42}$ differ from one another by only two amino acids, is clear that these two peptides differ significantly from one another at the structural level. The structural differences between these two peptides is reflected in their functional differences. The functional differences between $A\beta_{1-40}$ and $A\beta_{1-42}$ is discussed, for example, in Atwood *et al.*, *J. Neurochemistry* 75:1219-1233 (2000) (copy submitted herewith as Exhibit 9).

Moreover, the ability of persons of ordinary skill in the art to produce polyclonal antibodies that recognize $A\beta_{1-40}$ but not $A\beta_{1-42}$, and vice versa, (*see* Mak and the Oncogene catalog), demonstrates that these two peptides differ significantly at the structural level and that the overlap of these peptides at the amino acid sequence level would not have inhibited the production of such antibodies. Thus, the fact that the amino acid sequence of $A\beta_{1-40}$ is encompassed by the amino acid sequence of $A\beta_{1-42}$ does not suggest that it would have required undue experimentation to produce polyclonal antibodies that recognize $A\beta_{1-42}$ but not $A\beta_{1-40}$.

A person of ordinary skill in the art would have been able to produce polyclonal antibodies that are specific to $A\beta_{1-42}$ and do not cross-react with $A\beta_{1-40}$, and polyclonal antibodies that are specific to $A\beta_{1-40}$ and do not cross-react with $A\beta_{1-42}$, using only routine methods in the art. Therefore, Applicants respectfully request that the enablement rejection of claims 5-16 be reconsidered and withdrawn.

B. Written Description

Claims 5-16 were rejected under 35 U.S.C. § 112, first paragraph, as allegedly containing subject matter which was not described in the specification in such a way as to

reasonably convey to one skilled in the art that the inventors, at the time the application was filed, had possession of the claimed invention. (*See* Paper No. 20, page 7.) Applicants respectfully traverse this rejection.

This rejection is based on the assertion that "[t]he specification lacks adequate written description of the necessary immunogens and process steps to produce the polyclonal antibody . . . with the claimed specificities for use in the assay." (Paper No. 20, page 8.) This assertion assumes that the only way to produce polyclonal antibodies that are specific to $A\beta_{1-42}$ and that do not cross-react with $A\beta_{1-40}$, and vice versa, is by identifying particular immunogens that elicit the specific immune responses.

The identification of particular immunogens for producing the antibodies of the invention would have been a matter of routine experimentation in the art. (*See* Applicants' remarks set forth in the Supplemental Amendment and Reply, filed on January 10, 2003, pages 4-7.) For example, the specification sets forth the amino acid sequence of human Aβ peptide from which a skilled artisan could easily select immunogens for the production of $A\beta_{1-40}$- and $A\beta_{1-42}$-specific polyclonal antibodies. (*See, e.g.*, specification at page 23, lines 16-17, and Fig. 10.) Therefore, with respect to the identification of immunogens that can be used to produce the polyclonal antibodies, the written description requirement is fully satisfied.

In addition, as discussed above, the use of particular immunogens to produce polyclonal antibodies with desired specificities is only one of several methods that could have been used to produce the polyclonal antibodies that are used in the practice of method claims 5-16. (*See* remarks in Section IV.A., above.) Therefore, whether or not any specific

immunogens are recited in the specification, this assertion cannot properly form the basis of a written description rejection.

To satisfy the written description requirement of 35 USC § 112, first paragraph, an Applicant must convey with reasonable clarity to those skilled in the art that, as of the effective filing date, Applicants were in possession of the invention. *See Vas-Cath, Inc. v. Mahurkar*, 935 F.2d 1555, 1560, 19 USPQ2d 1111, 1117 (Fed. Cir. 1991). The present specification describes assays for detecting or quantifying Aβ peptide in a candidate solution. (*See* specification at page 5, line 20, through page 6, line 3.) The described assays involve the use of "an antibody specific for Aβ peptide." (*See* specification at page 5, line 30.) The specification indicates that "[t]he preferred antibodies used in the practice of the invention are those that are either specific to $A\beta_{1-42}$ which do not cross react with $A\beta_{1-40}$ or specific to $A\beta_{1-40}$ which do not cross react with $A\beta_{1-42}$." (*See* specification at page 7, lines 3-5.)

Prior to the effective filing date of the application, persons having ordinary skill in the art would have recognized that "antibodies specific to $A\beta_{1-42}$ which do not cross react with $A\beta_{1-40}$ or specific to $A\beta_{1-40}$ which do not cross react with $A\beta_{1-42}$" encompass both monoclonal and polyclonal antibodies with the recited specificities. Methods for producing and using polyclonal antibodies that recognize a particular peptide and do not cross-react with other closely related peptides were commonplace in the art. (*See* remarks in Section IV.A., above.) Therefore, persons of ordinary skill in the art, in view of the present specification, would have recognized that Applicants were in possession of the methods of claims 5-16. Accordingly, Applicants respectfully request that the written description rejection of claims 5-16 be reconsidered and withdrawn.

Conclusion

Applicants believe that a full and complete reply has been made to the outstanding Office Action. If the Examiner believes, for any reason, that personal communication will expedite prosecution of this application, the Examiner is invited to telephone the undersigned at the number provided.

Prompt and favorable consideration of this Reply is respectfully requested.

Respectfully submitted,

STERNE, KESSLER, GOLDSTEIN & FOX P.L.L.C.

Frank R. Cottingham
Attorney for Applicants
Registration No. 50,437

Date: July 7, 2003

1100 New York Avenue, N.W.
Washington, D.C. 20005-3934
(202) 371-2600

SKGF_DC1:147503.1